

IN THE CLAIMS

Please amend Claim 1 as follows, and cancel claim 13.

1. (Currently Amended) A time-resolved fluoroimmunoassay (TR-FIA) method for detecting a cytokine in a biological fluid sample, comprising:
subjecting the biological fluid sample to a heat treatment under non-denaturing temperature conditions for the cytokine; and

forming a composite by (a) binding a first antibody, including a portion bound to a solid phase and a region bindable to a cytokine, to a solid phase; (b) adding the sample containing the cytokine; (c) binding a second antibody, including a region bindable to the cytokine and a portion to which biotin is bound, to the cytokine; (d) adding a conjugate including streptoavidin or avidin and a fluorescent structural portion capable of being complexed with a lanthanoid metal ion; and (e) adding a the lanthanoid metal ion to bind the conjugate of (d) are bound, the composite being formed on the solid phase; and

measuring fluorescence of the fluorescent structural portion which has been complexed with the lanthanoid metal ion,

wherein the method comprises a step of washing after each of steps (a) to (c);
 and

wherein the cytokine is a cytokine belonging to the chemokine family, and

wherein the fluorescent structural portion is represented by General Formula (I):



(where R is a residue which is a functional group capable of forming a covalent bond with a protein; Ar is a hydrocarbon group having a conjugated double bond system; n is an integer equal to or greater than 1; and X is a fluorine atom or a group represented by General Formula (II):



2. (Original) A method according to claim 1, wherein the lanthanoid 1 metal ion is europium.

3. (Previously Presented) A method according to claim 1, wherein the fluorescent structural portion is represented by General Formula (III):



(where R, Ar, and n have the same definitions as in claim 1).

4. (Original) A method according to claim 3, wherein the fluorescent structural portion is 4,4'-bis(1", 1", 1", 2", 2", 3", 3"heptafluoro-4", 6"-hexanedion-6"-yl)-sulpho-oterphenyl.

5. (Original) A method according to claim 1, wherein 10 to 60 units of the fluorescent structural portion are present per molecule of streptoavidin or avidin in the conjugate.

6. (Original) A method according to claim 1, wherein the step of measuring fluorescence is performed without allowing the composite formed on the solid phase to dissociate.

7. (Original) A method according to claim 1, wherein the step of measuring fluorescence is performed after allowing the composite formed on the solid phase to dissociate.

8. (Canceled)

9. (Original) A method according to claim 1, wherein the cytokine is a CXC chemokine.

10. (Original) A method according to claim 9, wherein the cytokine is stromal cell-derived factor-1 (SDF-1).

11. (Original) A method according to claim 1, wherein the biological fluid sample is plasma or whole blood.

12. (Original) A method according to claim 1, further comprising, before the step of forming the composite, a step of diluting the biological fluid sample with a buffer solution used for sample dilution,

wherein the buffer solution used for sample dilution is 0.01 to 0.1 M tris-hydrochloric acid whose pH is 7.3 to about 8.3, the buffer solution containing 0.1 to 0.3% of bovine serum albumin, 0.05 to 0.2% of sodium azide, and 0.5 to 1.5% of sodium chloride.

13. (Canceled)

14. (Original) A method according to claim 1, further comprising, before the step of measuring fluorescence, a step of washing the composite formed on the solid phase with a buffer solution used for washing,

wherein the buffer solution used for washing the composite is 0.01 to 0.1 M tris-hydrochloric acid whose pH is 8.5 to about 9.5, the buffer solution containing 0.01 to 0.1% polyoxyethylenesorbitan monolaurate.

15. (Original) A method according to claim 1, wherein the solid phase is a microtiter plate having an IgG adsorption ability of 50 to 200 ng/cm².

16. (Previously Presented) A kit for a time-resolved fluoroimmunoassay (TR-FIA) method for detecting a cytokine in a biological fluid sample, comprising: a first antibody including a portion bound to a solid phase and a region bindable to a cytokine; a second antibody including a region bindable to the cytokine and a portion to which biotin is bound; a conjugate including streptoavidin or avidin and a fluorescent structural portion capable of being complexed with a lanthanoid metal ion; and the lanthanoid metal ion,

wherein the cytokine is a cytokine belonging to the chemokine family, and

wherein the fluorescent structural portion is represented by General Formula (I):



(where R is a residue which is a functional group capable of forming a covalent bond with a protein; Ar is a hydrocarbon group having a conjugated double bond system; n is an integer equal to or greater than 1; and X is a fluorine atom or a group represented by General Formula (II):

